

The study of physical properties of spray dried whey and milk permeates lactose

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Abstract. The aim of this study was to investigate substrate and environment effect on the physical state of lactose crystals, their stability and behaviour comparing with pure lactose which traditionally used in an analysis. Sweet and acid whey permeate as well as milk permeate were analysed. Mini spray-drier (BÜCHI B-290, Labortechnik AG, Switzerland) was used for the study. Lactose optical rotation was measured with a polarimeter, structural characteristic was carried out by X-ray diffractometer and glass transition analyses was made by TGA/DSC. α -Lactose monohydrate (Sigma-Aldrich, Germany) was used as a control. All spray-dried permeates samples showed amorphous state lactose crystals. The DSC analysis demonstrated a glass transition in the interval of 85–95 °C, melting 202–204 °C for spray-dried permeates lactose. In turn, the control sample showed crystallization at 158 ± 0.5 °C and a melting peak at 226 ± 0.5 °C. Optical rotation of spray-dried lactose obtained from sweet and acid whey permeate and milk permeate was in the range from 18 to 28°, control sample 52°. The study results showed that substrate, sample pH, ingredients and their derivatives impact lactose glass transition and mutarotation. The current study highlights the essential physical properties of spray-dried permeates lactose and the importance of its purity in food, cosmetic and pharmaceutical industry.

Key words: lactose, milk permeate, whey permeate, glass transition, mutarotation.

INTRODUCTION

Permeate is considered as a suitable substrate for lactose hydrolysis. The outcome of final sugars and its concentration depends on permeate type, solid content and medium environment. That gives a propose to analyse more closely properties and behaviours of lactose from permeate and build a new strategy to raise glucose-galactose syrup sweetness. Lactose is a disaccharide which consists of galactose and glucose. The main source of lactose is bovine milk containing approximately 4.8% carbohydrate (Jenab et al., 2018). Lactose is considering as a problem for whey utilization since it crystallizes at low temperatures. As one of the possibilities in food industry, lactose can hydrolyse by enzymatic or acid method (Das et al., 2015). Lactose is used for investigation and product formation in various biotechnological processes (Ryan & Walsh, 2016) obtaining lactose derivatives such as galacto-oligosacchrides, lactulose, lactitol, and lactobionic acid (Huppertz & Gazi, 2016). Whey can be fractionated into retentate (rich with protein) and permeate (rich with lactose) using ultrafiltration (Jenab et al., 2018).

The major compound in whey permeate and milk permeate is lactose in addition with a small number of soluble minerals and proteins. Whey permeates solids consist of 76 to 85% lactose, and 11 to 16% proteins and minerals, whereas milk permeate solid contains 78 to 88% lactose and 11 to 16% proteins and minerals (Pandalaneni & Amamcharla, 2018). In aqueous medium, lactose may exist in α and β forms but after drying process only anhydrous form. Crystallization of lactose is important in complex dairy systems under industrial process conditions, such as spray-drying, freeze-drying, and storage, such as ice-cream, evaporated milk and whey. In this case, the different crystals exist together with amorphous lactose and changing in overall the functional properties of lactose. α -Lactose monohydrate is the main lactose form in spray-dried lactose/whey permeate and lactose/gelatine mixtures (Gänzle et al., 2008). The most common method is spray-drying for dehydration of milk and whey products. During this process, moisture is removed and leads to the formation of lactose at amorphous state. Notable to highlight that the presence of protein(s), fat, minerals and lactic acid can largely influence the physical and chemical behaviours as well as water absorption, glass transition temperature and crystallization of the lactose (Shrestha et al., 2007). In particular, the focus of the study was to analyse the spray-dried sweet and acid whey, as well as milk permeates powders to gain the useful information of powders physical properties which could be taken in consideration as a prospective information in food products production and physical properties assessment. The aim of this study was to investigate substrate and environment effect on lactose crystals, their stability and behaviour comparing with pure lactose which traditionally used in an analysis.

MATERIALS AND METHODS

Materials

Acid whey and milk permeate were kindly donated by JSC 'Tukuma piens'. Sweet whey was derived from dairy Ltd 'Latvijas Piens' where further ultrafiltration was done in the laboratories of Latvia University of Life Sciences and Technologies. α -Lactose monohydrate $\geq 99\%$ as a control was purchased from Sigma-Aldrich (Latvia).

Preparation of permeate

Sweet whey was treated using cross-flow membrane filtration Armfield FT17 (UK) and polymer membrane filter GKSP, 92 mm, Sterlitech (USA) with molecular weight cut-off of 3 kDa was used in the study, all process was operated at temperature 4 ± 2 °C and pressure 28 ± 2 bar. Sweet whey was stored in the fridge. Acid whey and milk permeates were produced on an industrial scale using ultrafiltration UF Unit, GEA (Germany) equipped with spiral membranes. Sweet, acid and milk permeate was analysed by MilkoScanTM Mars, Foss Analytical A/S (Denmark) for lactose, protein and fat, total solids content determination and pH measurement.

Preparation of spray-dried lactose and permeates powders

To obtain a powder from sweet and acid whey as well as milk permeate and 5% w/v aqueous α -lactose monohydrate solution was used BÜCHI mini spray-drier B-290 (Labortechnik AG (Switzerland)), experiment was done based on Chandrapala & Vasiljevic (2017), also Islam & Langrish (2010) works with some modifications. To objectively analyse the impact of permeate origin on lactose physical and chemical

parameters under the same conditions was spray-dried 5% w/v aqueous α -lactose monohydrate solution (initial material α -lactose monohydrate purity $\geq 99\%$) and compared with spray-dried permeate powders lactose.

The spray-dryer was used with following conditions: aspirator rate 100%, the flow rate of the feed solution 40–50 mL min⁻¹, an inlet air temperature 170 °C and an outlet temperature 115–120 °C. The collected powders were immediately placed in 50 mL tube and stored in a desiccator with 0–3% relative humidity at room temperature till further analysis.

Instrumental analysis

Optical rotation

Polarimeter Polax-2L, Atago (USA) was used for measurement of lactose optical rotation. Each sample was dissolved in deionized water at a concentration of 1% (w/v) and placed into 100 mm cuvette. To calculate sugar optical rotation, the following equation was used:

$$[\alpha]D = \frac{\alpha}{I \cdot C} \quad (1)$$

where I – length of cuvette, dm; C – the sample concentration in g 100 mL⁻¹ (Chandrapala & Vasiljevic, 2017).

X-ray powder diffraction

Structural characterization of samples was carried out using diffractometer Bruker AXS, D8 Advance (Germany) with CuK α 1 radiation at $\lambda = 1.5418$ Å and a position sensitive detector (PSD). The tube was operated at voltage 40 kV and current 40 mA. Scan range 5°–60° on 2 θ scale at rate 5° min⁻¹ was used according to Wu et al. (2014) method. The database ICDD PDF2 was used for patterns analysis.

Differential scanning calorimetry (DSC) and thermogravimetric analysis (TGA)

Analyses were performed on a Mettler Toledo TGA/DSC (Switzerland) operating with the STARE System Software. In one sample measurement, instrument was performing thermal transition and thermogravimetric analysis. Sample 5–10 mg was weighted into an aluminium pan and heated at the temperature range 30 to 300 °C within heating rate of 10 °C min⁻¹. An empty aluminium pan was used as a reference in every test (Badal Tejedor et al., 2018; Veldre et al., 2011).

Scanning Electron Microscopy (SEM)

The morphology of the lactose crystal samples was examined using scanning electron microscopy SEM-FIB Tescan Lyra with an accelerating voltage of 12 kV. Each sample was placed onto a carbon tape on an aluminium sample disc and a compressed gas was used to remove unfixed powder particles. Lactose samples were coated with a 27 nm gold layer using Quorum Q150R coating unit at 25 mA for 45 s (Kougoulos et al., 2010).

Data analysis

Results were expressed as mean \pm standard deviation (SD) of three replicates for composition measurements. Statistical analyses were used with analysis of *One-Way ANOVA* and *Tukey* test at the significance level $P < 0.05$.

RESULTS AND DISCUSSION

Ultrafiltration was used to remove low molecular weight components but keep lactose in the same concentration. Table 1 represents the amount of main components and medium pH of each permeate before spray-drying.

Table 1. Proximate composition and pH of permeates before spray-drying

Permeate	Fat, %	Proteins, %	Lactose, %	Total solid, %	pH
Sweet whey	0 ^a	0.2 \pm 0.1 ^a	3.8 \pm 0.1 ^a	4.6 \pm 0.1 ^a	6.1 \pm 0.1 ^a
Acid whey	0 ^a	0.5 \pm 0.1 ^b	4.2 \pm 0.2 ^b	5.2 \pm 0.2 ^b	4.6 \pm 0.1 ^b
Milk	0 ^a	0.5 \pm 0.1 ^b	4.7 \pm 0.1 ^c	6.0 \pm 0.1 ^c	5.8 \pm 0.1 ^c

Results indicated with the same letter within a column do not differ significantly ($P > 0.05$).

Permeate composition varies depending on initial product for ultrafiltration and the condition of ultrafiltration in terms of membrane type, concentration rate and other factors (Barile et al., 2009). Sweet whey storage conditions could activate lactose crystallisation gaining molecular weight therefore part of sugar during ultrafiltration was not able to get through membranes as it showed in Table 1. Parameters of each membrane also affected the condition of what size of the particle was removed. Besides of minimal amount of protein, permeate solid contains also mineral salts. Salts are considering as an important factor which associates with lactose and affects the crystallisation of spray-dried lactose. The effect of salts differs with their types and amount (Imtiaz-Ul-Islam & Langrish, 2008). This should be taken into consideration if spray-dried lactose and permeate powder intended to use in enzymatic lactose hydrolysis. The ionic environment affects enzymatic activity and evolution of lactose hydrolysis reaction (Demirhan et al., 2008).

Sugar optical rotation $[\alpha]_D$ measurements (Table 2) showed that $[\alpha]_D$ of α -lactose monohydrate was $52 \pm 0.1^\circ$ and its corresponding to the information on sample specification provided by producer. All spray-dried samples had lower $[\alpha]_D$ than α -lactose monohydrate but significant difference was not observed among powder from acid whey and milk permeates as well as 5% (w/v) lactose solution. The low $[\alpha]_D$ of sweet whey permeate powder could be related to the solid content which left during ultrafiltration.

Table 2. Optical rotation of α -lactose monohydrate (control) and lactose in spray-dried permeates powders

Samples	Optical rotation, ($^\circ$)
α -lactose monohydrate	52 \pm 0.1 ^a
Spray-dried:	
5% (w/v) lactose solution	34 \pm 0.1 ^b
Sweet whey permeate	20 \pm 0.3 ^c
Acid whey permeate	30 \pm 0.3 ^b
Milk permeate	27 \pm 0.2 ^b

Results indicated with the same letter within a column do not differ significantly ($P > 0.05$).

The mutarotation rate of lactose is greatly affected by spray-drying temperature and pH as well as presence of other sugars and salts reported by Huppertz & Gazi (2016). During spray-drying in a small part of time substrate was heated up till 170 °C and outlet from a nozzle at 115–120 °C that may consider as one of the factors which caused the lactose transformation as well as intensive mutarotation between α and β lactose (Hynes & Zalazar, 2008).

The morphology of lactose crystals in spray-dried permeates powder samples and α -lactose monohydrate crystals is showed in Fig. 2.

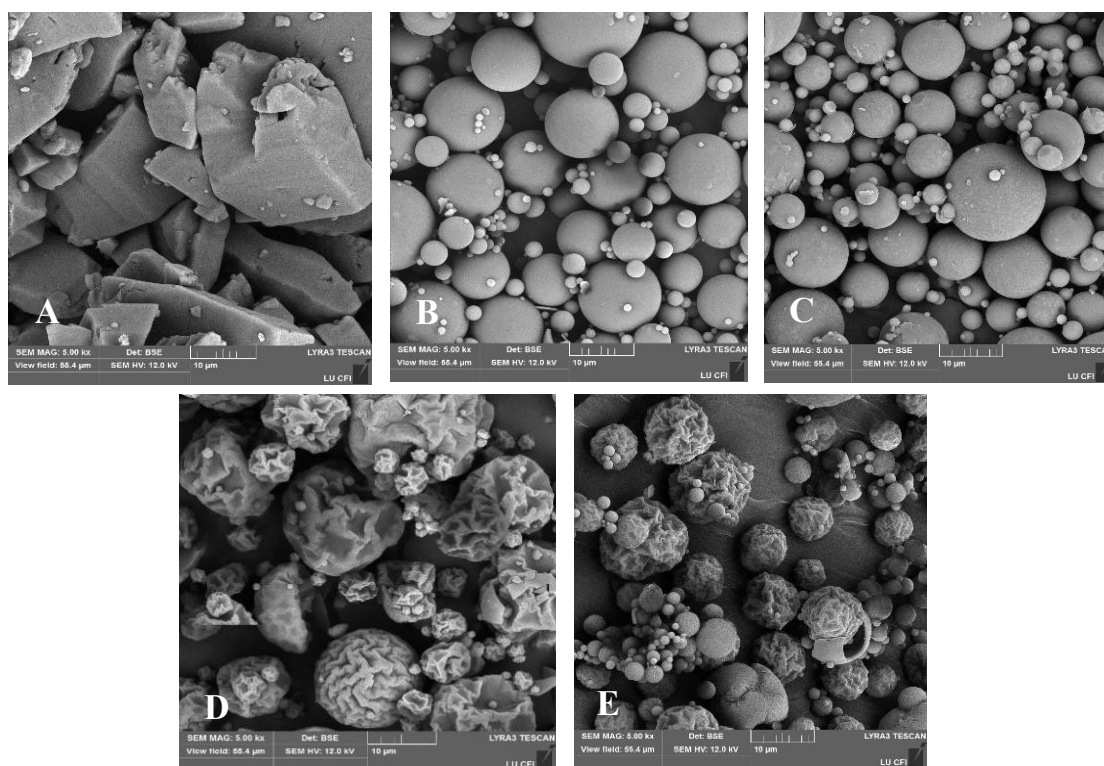


Figure 2. SEM surface morphology of α -lactose monohydrate crystals (A) and spray dried lactose obtained from 5% lactose solution (B); from sweet whey permeate (C); from acid whey permeate (D); from milk permeate (E). All micrographs are shown at magnification 5,000 \times .

Fig. 2, A shows the typical crystal shapes of α -lactose monohydrate, such as diamond-shaped plate and pyramids. Spray-dried lactose powder and permeate powders have an amorphous lactose form and can be observed in Fig. 2, B–E. During spray-drying of substrates, water evaporation and substrate viscosity increase so fast that crystal formation cannot happen and lactose forms into amorphous form (Gänzle et al., 2008). In Fig. 2, B is a standard crystalline form in a fresh spray-dried powder. According to the findings in the literature spray-dried conditions have the significant impact on powder properties, such as initial product solid content, drying temperature, initial droplet size and others (Kim et al., 2009). In this study, the appearance of droplets obtained from different permeate powders was not similar Fig. 2, B–E. Droplet surface of spray-dried lactose obtained from 5% lactose solution Fig. 2, B and from sweet whey permeate Fig. 2, C was spherical in shape with a smooth surface and the size of particles in both figures was relatively similar. However, the surface of the spray-dried droplets

obtained from acid whey Fig. 2, D and milk Fig. 2, C has various shapes. One part of droplets has shrivelled appearance, another part has fissures or breakages and another part was smooth. This can be explained by the findings of Kim et al. (2009) where the solid content and drying temperature affected droplet surface and coverage. These observations are confirmed by the results in Table 1, which clearly shows the significant difference of the total solids content among permeates as well as pH. Acid whey and milk permeates in Table 1 showed that pH was below 6 and might affected more strongly the surface appearance of droplets. Initial product solid content (fat, protein, salts, lactose) and pH are responsible for the dried droplet surface formation. Depending on content and pH in permeate during spray-drying, substances are interacting with each other that reflects on the surface, particles having lactose coverage and lactose crystallinity (Ebrahimi et al., 2015).

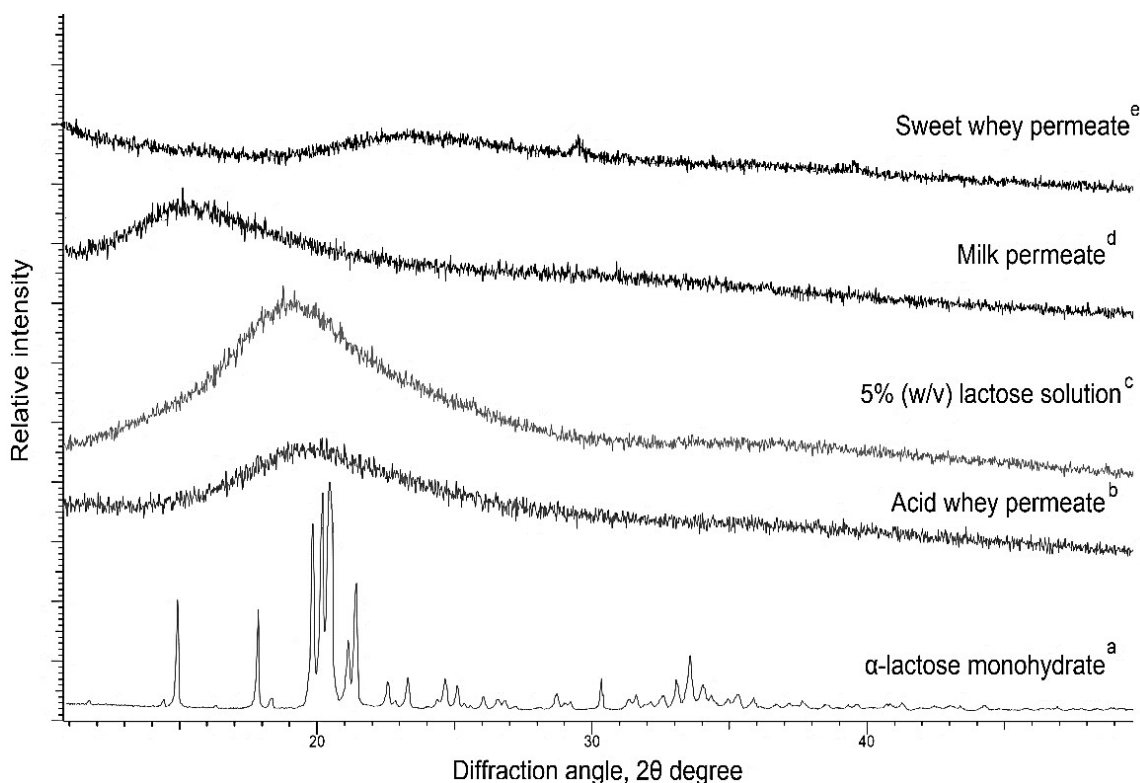


Figure 3. X-ray diffraction patterns of α -lactose monohydrate crystals and spray-dried lactose obtained from 5% (w/v) lactose solution; sweet whey permeate; acid whey permeate; milk permeate. Results indicated with the different letter differ significantly ($P < 0.05$).

The X-ray analysis was used to determine the crystalline form of commercial α -lactose monohydrate and spray-dried lactose from different substrates. Crystal properties of all samples were observed in Fig. 3. All patterns of spray-dried powders approved that using particular spray-drying conditions lactose obtained an amorphous (non-crystalline) form. It was not possible to identify the polymorphs in these certain patterns. To the same observations came up Price & Young (2004) in the study was used inlet temperature 185 °C and pattern of spray-dried lactose was without any major crystalline long-sharp peak instead of α -lactose monohydrate. The database ICDD PDF2

identified in pattern of spray-dried sweet whey permeate powder the presence of potassium chloride at diffraction angle $28 \pm 0.5^\circ$ and $40 \pm 0.5^\circ$ 2θ degree. That could be related to the interaction between lactose and this particular salt under certain conditions. The pattern differences might depend on the composition and amount of elements in each substrate, drying temperature and medium pH (Chen et al., 2015). In contrast, α -lactose monohydrate showed sharp diffraction peaks which allowed to identify the types of lactose crystal forms. In product, specification was mentioned the presence of β -lactose which amount is so less that it was difficult to identify this crystal.

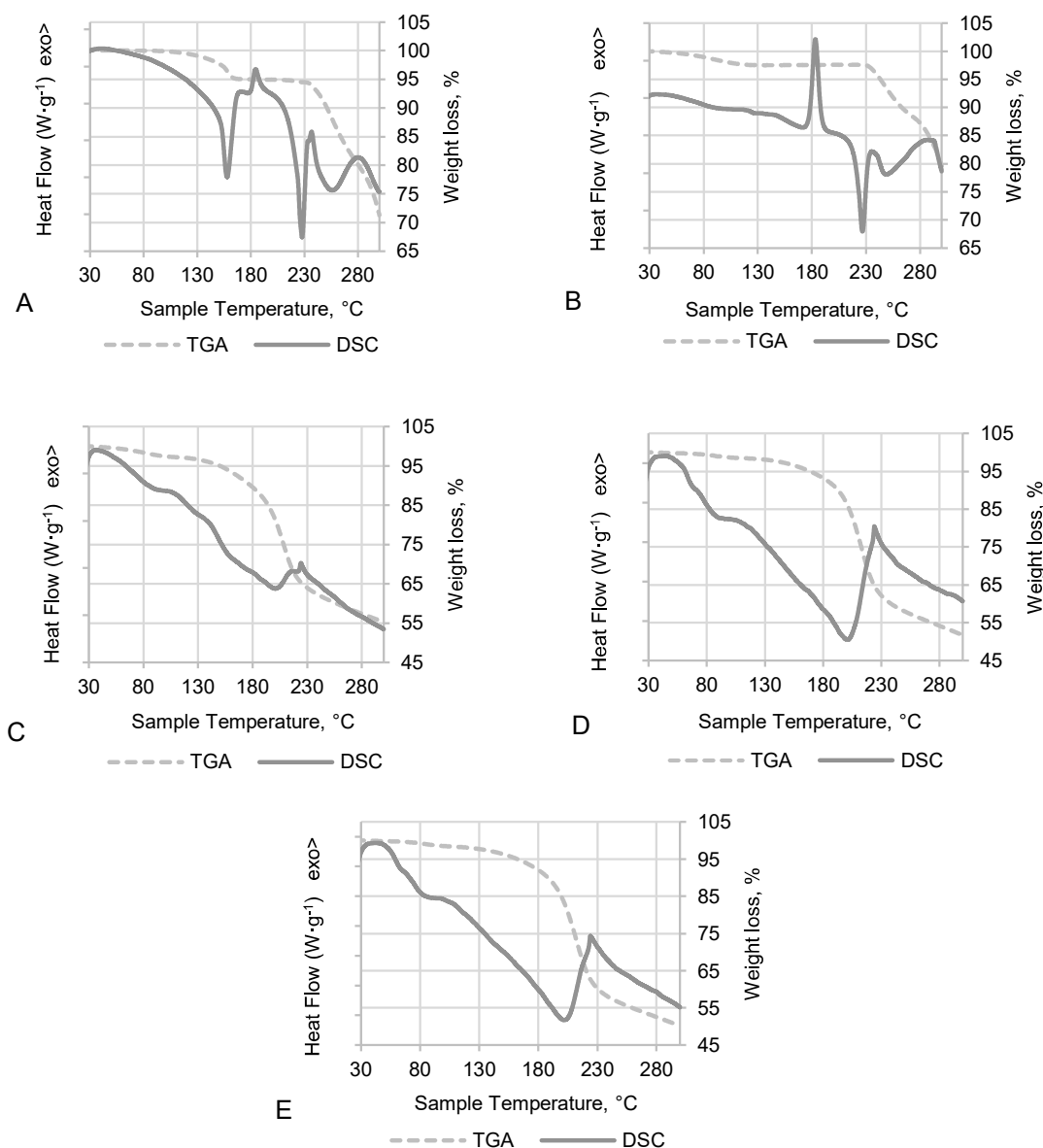


Figure 4. DSC thermal profile and TGA weight loss profile for α -lactose monohydrate crystals (A) and spray-dried lactose obtained from 5% lactose solution (B); from sweet whey permeate (C); from acid whey permeate (D); from milk permeate (E).

The crystallinity was analysed by measuring thermal properties of the sample upon heating using DSC and the loss of moisture by TGA. Both profiles of the lactose samples are showed in Fig. 4. Samples were heated at the temperature range from 30 to 300 °C showing the difference among samples. The two endothermic peaks were showed in DSC profile of α -lactose monohydrate (see Fig. 4, A). The first peak at 158 ± 0.5 °C expressed the evaporation of the crystallized water, the second peak at 226 ± 0.5 °C showed that lactose sample melted before decomposition and the total weight loss was $27.5 \pm 0.5\%$. The amorphous spray-dried lactose Fig. 4 B glass transition started at 58 ± 1 °C where weight loss was $2 \pm 0.5\%$, recrystallization started at 183.5 ± 0.5 °C and melting peak at 227 ± 0.5 °C where also occurred a rapid weight loss of $18 \pm 1\%$. It should be noted that α -lactose monohydrate crystal contains approximately 5% of water and water is less strongly bounded. During recrystallization at 184 ± 1 °C α -lactose monohydrate (Fig. 4, A) and amorphous spray-dried lactose (Fig. 4, B) according to Badal Tejedor et al. (2018) study converted into the same physical form which led to the same melting temperature. Lactose transformation into α and β forms during thermal treatment are not excluded. The DSC profiles of spray-dried permeate powders Fig. 4, C–E were similar ($F_{\text{crit}} = 5.14$, $P > 0.05$). The glass transition to permeate samples Fig. 4, C–E started in the interval of 85–95 °C and melting of samples were observed at 202 ± 2 °C. During the melting phase the weight loss to lactose samples from permeates Fig. 4, C–E were in the range of 30–45%. Results indicate that presence of other substances such as fat, protein, salts, lactic acid and medium environment decrease lactose thermal resistance. The DSC profiles highlight the physical properties and the differences between lactose from permeates and pure lactose.

CONCLUSIONS

The present study showed that the physical properties of spray-dried permeate powders are strongly influenced by the presentence of substances and condition. Lactose is very sensitive sugar and environment conditions can easily affect its behaviour. Study results reveals permeate powders capability of lactose crystal transformation and thermal transition behaviour where each of powder showed slightly different results. Research showed that permeate powders do not contain highly pure lactose, also the presence of other substances is established which effect their physical properties. Knowing lactose physical properties in permeate powders, they could help to avoid and prevent difficulties during food processing and storage, as well as in permeates quality evaluation.

Further studies are necessary to evaluate spray-dried permeate powders as an option in the food industry to use their as a substrate in enzymatic lactose hydrolysis. Permeate powders contain mineral salts which can act as an activators for β -galactosidases and contribute the hydrolysis reaction.

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